

Attorney Docket No.: ISPH-0613  
Inventors: Crooke et al.  
Serial No.: 10/054,313  
Filing Date: October 22, 2001  
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Listing of Claims:

Claim 1 (original): An isolated human RNase polypeptide comprising human Type 2 RNase H.

Claim 2 (original): The isolated human RNase polypeptide of claim 1 wherein the polypeptide comprises SEQ ID NO: 1.

Claim 3 (original): An isolated human RNase polypeptide prepared from a culture of ATCC Deposit No. 98536.

Claim 4 (original): A cloned and expressed human RNase H polypeptide.

Claim 5 (original): The cloned and expressed human RNase H polypeptide of claim 4 which is a human Type 2 RNase H polypeptide.

Claim 6 (original): The cloned and expressed human RNase H polypeptide of claim 4 which is a human RNase H1 polypeptide.

Claim 7 (original): The cloned and expressed human RNase H polypeptide of claim 4 which comprises SEQ ID NO: 1.

Claim 8 (original): The cloned and expressed human RNase H polypeptide of claim 4 which is prepared from a culture of ATCC Deposit No. 98536.

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Claim 9 (original): A composition comprising a cloned and expressed human RNase H polypeptide and a pharmaceutically acceptable carrier.

Claim 10 (original): The composition of claim 9 wherein the human RNase H polypeptide is a human Type 2 RNase H polypeptide.

Claim 11 (original): The composition of claim 9 wherein the human RNase H polypeptide is a human RNase H1 polypeptide.

Claim 12 (original): A composition comprising a human RNase H polypeptide and a pharmaceutically acceptable carrier.

Claim 13 (original): The composition of claim 12 further comprising an antisense oligonucleotide, wherein the human RNase H polypeptide is a human Type 2 polypeptide.

Claim 14 (original): An isolated polynucleotide encoding a human RNase H polypeptide.

Claim 15 (original): The isolated polynucleotide of claim 14 which is a human Type 2 RNase H.

Claim 16 (original): A vector comprising a nucleic acid encoding a human RNase H polypeptide.

Claim 17 (original): A host cell comprising the vector of claim 16.

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Claim 18 (original): A composition comprising a vector comprising a nucleic acid encoding a human RNase H polypeptide and a pharmaceutically acceptable carrier.

Claim 19 (original): The composition of claim 18 further comprising an antisense oligonucleotide, wherein the human RNase H polypeptide is a human Type 2 RNase H polypeptide.

Claim 20 (original): An antibody targeted to a human Type 2 RNase H polypeptide.

Claim 21 (original): A nucleic acid probe capable of hybridizing to a portion of a nucleic acid encoding a human Type 2 RNase H polypeptide.

Claim 22 (original): A human Type 2 RNase H--his-tag fusion polypeptide.

Claim 23 (original): An antisense oligonucleotide capable of eliciting cleavage of its complementary target RNA by a human Type 2 RNase H polypeptide wherein said human Type 2 RNase H polypeptide comprises SEQ ID NO: 1.

Claim 24 (original): A method of enhancing inhibition of expression of a selected protein by an antisense oligonucleotide targeted to an RNA encoding the selected protein comprising:

(a) providing an antisense oligonucleotide targeted to an

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RNA encoding a selected protein whose expression is to be inhibited;

(b) allowing said oligonucleotide and said RNA to hybridize to form an oligonucleotide-RNA duplex;

(c) contacting said oligonucleotide-RNA duplex with a human Type 2 RNase H polypeptide, under conditions in which

cleavage of the RNA strand of the oligonucleotide-RNA duplex occurs,

whereby inhibition of expression of the selected protein is enhanced.

Claim 25 (original): The method of claim 24 wherein the human Type 2 RNase H polypeptide comprises SEQ ID NO: 1.

Claim 26 (original): The method of claim 25 wherein the antisense oligonucleotide is a chimeric oligonucleotide.

Claim 27 (original): A method of screening oligonucleotides to identify an effective antisense oligonucleotide for inhibition of expression of a selected target protein comprising:

(a) contacting a human Type 2 RNase H polypeptide with an RNA encoding the selected target protein and an oligonucleotide complementary to at least a portion of the RNA under conditions in which an oligonucleotide-RNA duplex is formed;

(b) detecting cleavage of the RNA of the oligonucleotide-RNA duplex wherein cleavage is indicative of antisense efficacy.

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Claim 28 (original): The method of claim 27 wherein the human Type 2 RNase H polypeptide is enriched or overexpressed.

Claim 29 (original): The method of claim 27 wherein the human Type 2 RNase H polypeptide is exogenously added.

Claim 30 (original): The method of claim 27 wherein the human Type 2 RNase H polypeptide is an isolated, purified human Type 2 RNase H polypeptide.

Claim 31 (original): An effective antisense oligonucleotide identified in accordance with the method of claim 27.

Claim 32 (original): The method of claim 27 further comprising determining the site on the RNA at which cleavage occurs, whereby said site is identified as a Type 2 RNase H-sensitive site.

Claim 33 (original): The method of claim 32 further comprising identifying an effective antisense oligonucleotide which hybridizes to said Type 2 RNase H-sensitive site.

Claim 34 (original): The method of claim 27 wherein the oligonucleotide is one of a mixture or library of oligonucleotides.

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Claim 35 (original): An effective antisense oligonucleotide identified in accordance with the method of claim 33.

Claim 36 (original): A method of making an antisense oligonucleotide which elicits cleavage of its complementary target RNA by a human Type 2 RNase H polypeptide comprising synthesizing an oligonucleotide which is targeted to a selected RNA wherein said oligonucleotide, when hybridized to the selected RNA target to form a duplex, will bind the human

Type 2 RNase H polypeptide which thereby cleaves the RNA strand of the duplex.

Claim 37 (original): A method of prognosticating efficacy of antisense therapy of a selected disease comprising measuring the level or activity of a human Type 2 RNase H in a target cell of the antisense therapy.

Claim 38 (original): A method of identifying agents which increase or decrease activity of levels of a human RNase H polypeptide in a host cell comprising:

(a) contacting a cell expressing a human RNase H polypeptide with an agent suspected or increasing or decreasing activity or levels of the human RNase H polypeptide; and

(b) measuring the activity or levels of the human RNase H polypeptide in the presence and absence of the agent so that an increase or decrease in the activity or levels of the human RNase H polypeptide can be determined.

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Claim 39 (original): A method of identifying agents which increase or decrease activity or levels of an RNase H polypeptide comprising:

a) contacting an RNase H polypeptide with an agent suspected of increasing or decreasing activity or levels of said RNase H polypeptide.

b) measuring the activity or levels of the RNase H polypeptide in the presence and absence of the agent so that an increase or decrease in the activity or levels of the human RNase H polypeptide can be determined.

Claim 40 (original): The method of claim 39 wherein the RNase H polypeptide is a cloned and expressed RNase H polypeptide.

Claim 41 (original): The method of claim 39 wherein the RNase H polypeptide is a human RNase H polypeptide.

Claim 42 (original): The method of claim 39 wherein the RNase H polypeptide is a human RNase H polypeptide having SEQ ID NO: 1.

Claim 43 (original): The method of claim 39 wherein the RNase H polypeptide is prepared from a culture of ATCC Deposit No. 98536.

Claim 44 (original): A method of making substantially pure human Type 2 RNase H comprising transfecting a host cell with a vector containing a nucleic acid sequence encoding human Type 2

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RNase H, wherein said host cells express the human Type 2 RNase H polypeptide, and isolating the human Type 2 RNase H polypeptide.

Claim 45 (original): The method of claim 44 wherein said human Type 2 RNase polypeptide comprises SEQ ID NO: 1.